Claims 43-48 stand rejected for lack of novelty over Allen et al. Applicant has previously pointed out that Allen et al. provides no teaching or suggestion of any composition that is substantially free of myotubes and/or that contains at least 5% fibroblasts, as recited in the present claims. The Examiner has maintained the rejection, and has pointed to various sections of Allen et al. for support. Applicant respectfully submits that Allen et al. does not contain the disclosure indicated by the Examiner.

With respect to a lack of myotubes, the Examiner points to a section of Allen et al. (page 526, second column) that describes cultures containing BrdU. It is true that these cultures did not contain myotubes. However, they also did not contain "isolated skeletal myoblasts". Skeletal myoblasts, as defined in the present specification and as understood in the art, are cells that are "precursors of myotubes and skeletal muscle fibers"[1]. Such cells are defined by a capacity to differentiate into myofibers[2]. The present specification includes extensive discussion of preferred compositions being able to undergo, and/or having undergone, a specified number of population doublings[3]. The BrdU-containing compositions described by Allen et al. *cannot* differentiate and therefore are not compositions of isolated skeletal myoblasts. BrdU is an analog of thymidine that gets incorporated into the DNA of replicating cells. For a variety of different cell types, incorporation of BrdU into a cell's genome blocks the ability of the cell to differentiate[4]. Specifically for myogenic cells, it has been shown that incorporation of BrdU in a mouse myoblast line blocks myogenic differentiation[5]. The data presented by Allen et al. provide further evidence of this effect: identical cells under otherwise identical conditions divide, differentiate, and fuse to form myotubes in the absence of BrdU. Allen et al. added BrdU for the purpose of inhibiting differentiation[6]. Thus, the cells described by Allen et al. that had BrdU incorporated into their genomes were incapable of differentiating into myotubes and muscle fibers and therefore were not "skeletal myoblasts" according to the present claims.

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[1] See, for example, page 5, lines 19-20, which read "The terms 'skeletal myoblasts' and 'skeletal myoblast cells' are used interchangeably herein and refer to a *precursor of myotubes and skeletal muscle fibers*", emphasis added

[2] See, for example, page 5, lines 20-23, which read "The term 'skeletal myoblasts' also includes satellite cells .... Sattelite cells lie near the basal lamina of skeletal muscle myofibers and *can differentiate into myofibers*", emphasis added.

[3] See, for example, page 8, line 27-page 9, line 10.

[4] See, for example, Rutter et al., *Annu Rev Biochem* 42:601, 1973; Stockdale et al., *J. Cell Biol.* 44:134, 1970; Weintraub et al., *J. Mol. Biol.* 70:337, 1972; Abbot et al., *Proc. Natl. Acad. Sci. USA* 59:1144, 1968; Sax et al., *J. Bio. Chem.* 260:3812, 1985).

[5] Tapscott et al., *Science* 245:532, 1989.

[6] See page 526, column 2, lines 3-6, which read "In some experiments, *differentiation was inhibited . . . by including BrdU in culture medium*", emphasis added.

With respect to the presence of at least 5% fibroblasts (e.g., claim 44), particularly in compositions that had been cultured for at least 7 days (e.g., claim 46), Applicant previously pointed out that the only cultures grown by Allen et al. for such time periods were clones of satellite cells. In the Final Office Action, the Examiner does not dispute Applicant's position, but states "said 'clones' are taught by Allen to be low density compositions of satellite cells which absent evidence to the contrary contain fibroblast cells". Applicant respectfully submits that the "evidence to the contrary" is that the composition described by Allen et al. is a *clone*. A *clone* is a colony of cells obtained from a *single cell*. A *clone* of satellite cells cannot include any fibroblasts. Allen et al. did not observe any colonies that contained both cells that stain for desmin (e.g., myoblasts) and cells that do not (e.g., fibroblasts) (see page 530, column 1, which reads "Rat satellite cell colonies containing both stained and unstained cells were not found"). Similarly, the colonies that contained fibroblasts did not have any myoblasts (see page 530, column 1, lines 4-5, which reads refers to "large fibroblast colonies", and states that "there was no evidence of desmin staining in cells within these [fibroblast] cells" [page 530, column 1, lines 4-5]).

In light of all of these remarks, Applicant respectfully requests that the Examiner reconsider and remove the rejection for lack of novelty.

Rejection for Lack of Written Description/New Matter

The Examiner has rejected all claims on the ground that the current claims are directed to a subgenus of compositions (i.e., those lacking myotubes and containing not less than 5% fibroblasts) that was not sufficiently supported in the specification as filed.

Applicant respectfully disagrees. With respect to the "substantially free of myotubes" limitation, Applicant challenges the assertion that this language specifies a subgenus of the compositions described in the application. The phrase "substantially free of myotubes" simply specifies that substantially all of the skeletal myoblasts in the claimed composition are still in myoblast form. The entire specification is dedicated to describing compositions that, when implanted into cardiac tissue, will engraft and form myotubes. The compositions must contain myoblasts (rather than myotubes) at the time of delivery; if myotubes have already formed, they will not be able to form *in situ* and engraftment will not occur. The application is clearly

directed to cellular compositions containing myoblasts alone or with other non-muscle cells (e.g., fibroblasts); such compositions would all be substantially free of myotubes.

Even if the phrase "substantially free of myotubes" does refer to a subgenus of the skeletal-myoblast-containing compositions in the specification, it is a subgenus that is thoroughly described. The application makes clear that maturation of muscle cells in the inventive compositions should desirably be avoided. For instance, the specification contains a long paragraph specifying the desirability of *limited* doublings, and therefore limited cell differentiation/maturation[7]. The specification also specifically lists preferred percentages of skeletal myoblasts that should desirably be present in inventive compositions[8]. Several of these percentages are above 90%; 95%, 96%, 97%, 98%, and 99% are each specifically recited. Clearly, a composition that is 99% skeletal myoblasts is substantially free of myotubes. The specification makes clear that preferred compositions contain only skeletal myoblasts and fibroblasts[9], so that compositions containing skeletal myoblasts and fibroblasts, and substantially free of myotubes, are fully described in and supported by the specification.

The Examiner has also rejected claim 49 as new matter on the ground that "there is not a single exemplification of said composition". Applicant respectfully points out that a particular embodiment of an invention need not be presented in the Examples to be fully described in a specification. The present specification makes clear (at many points) that preferred compositions are compositions of human cells. For example, the specification states "The cells used in this invention can be derived from a suitable mammalian source, e.g., pigs *or from humans* . . . . In preferred embodiments, the cells are *human cells* . . . " (emphasis added; page 8, lines 9-11). See also page 9, lines 3-5, which points out that the optimal doubling numbers presented in the specification are for human cells.

The Examiner further argues that because, as Applicant has previously pointed out, the techniques used by Allen et al. give different results with cells of different origins, the present specification fails the written description requirement for claims to human cells because Examples 4-6 present data on rat cells. This argument is flawed.

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[7] See, for example, page 8, line 27-page 9, line 10.

[8]. See, for example, page 9, lines 13-15, which read "In another embodiment the composition comprises at least about 20%, 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, or 99% myoblasts"

[9] See, for example, page 9, lines 23-page 10, line 7.

First of all, Allen et al.'s work is completely different from the work of the present inventors; a lack of species generality in Allen et al.'s work cannot and should not be transferred to the present inventive work. The specification indicates, and as Applicant explained in person in an Interview held at the Patent and Trademark Office on January 15, 2003, that the techniques detailed in the present specification are equally applicable to cells of various origins. Furthermore, it is simply incorrect to assert that the specification does not contain exemplification of human myoblast compositions. In fact, in addition to the rat cell work exemplified in Examples 4-6, the specification includes Examples of work done with dog myoblasts (see, for example, Example 2) and of work done with human myoblasts (see, for example, Example 1, which begins on page 34, line 26; see in particular page 35, lines 9-10, which read "Thus, in dogs HM [human myoblasts] can be implanted and survive in the periphery of infarcted myocardium").

In support of these points, Applicant submits the enclosed Declaration by Dr. Jonathan Dinsmore, attesting that 1) a person of ordinary skill in the art, reading the present specification, would understand it to describe cell populations that are substantially free of myotubes; 2) a person of ordinary skill in the art, reading the present specification, would understand it to describe preparations of human cells, and particularly to describe preparations that are substantially free of myotubes; and 3) the preparations of human cells described in the present specification are in fact substantially free of myotubes.

For all of these reasons, Applicant respectfully submits that the rejections pending in the present case should be removed, and the application should pass to issuance.

It is Applicant's understanding that there are no fees associated with this matter. Should this understanding be in error, please charge any fees to our Deposit Account No. 03-1721.

Respectfully submitted,

Dated: 5/27/03

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Appendix
Pending Claims

43. A composition comprising:
isolated skeletal myoblasts; and
isolated fibroblast cells,
the composition being substantially free of myotubes.

44. The composition of claim 43, which composition comprises at least 5% fibroblasts.

45. The composition of claim 43 or claim 44, comprising:
isolated human skeletal myoblasts; and
isolated human fibroblasts,
the composition being substantially free of myotubes.

46. The composition of claim 43 or claim 44, wherein the composition is cultured *in vitro* for at least 7 days.

47. The composition of claim 43 or claim 44, which composition comprises cells that have been cultured *in vitro* for fewer than 20 population doublings.

48. The composition of claim 43 or claim 44, which composition comprises a collection of isolated cells consisting essentially of skeletal myoblasts and fibroblasts.

49. The composition of claim 48, which composition comprises a collection of isolated cells consisting essentially of human skeletal myoblasts and human fibroblasts.